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GB Virus C/Hepatitis G Virus Infection in KwaZulu Natal, South Africa

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Sera from 70 patients on maintenance haemodialysis, 98 patients with chronic liver disease, and 232 volunteer blood donors in the province of KwaZulu Natal, South Africa, were screened for GB virus/hepatitis G virus (GBV-C/HGV) RNA and anti-E2 by reverse transcription-polymerase chain reaction (RT-PCR) and by an enzyme-linked immunosorbent assay (ELISA), respectively. GBV-C/HGV RNA was detected in 17/70 (24.3%) haemodialysis patients, 12/98(12.2%) patients with chronic liver disease, and 44/232 (18.9%) blood donors (Africans [29/76; 38.2%]; Asians [2/52; 3.8%]; Whites [11/49; 22.4%], and "Coloureds" [persons of mixed origin; 2/55; 3.6%]). Overall (anti-E2 and/or RNA) 43.9% (43/98) of patients with chronic liver disease, 47.1% (33/70) of haemodialysis patients, and 31.9% (74/232) of blood donors (Africans [44/76; 5.9%]; Asians [5/52; 9.6%]; Whites [15/49; 30.6%], and Coloureds [9/54; 16.6%]) were exposed to GBV-C/HGV infection. There was a significant difference in the prevalence of GBV-C/HGV infection (RNA and/or anti- E2) between African blood donors and the other racial groups ($P < .001$), and between blood donors and haemodialysis patients ($P = .02$) and patients with chronic liver disease ($P = .04$). Anti-E2 antibodies and GBV-C/HGV RNA were almost mutually exclusive. GBV-C/HGV-infected haemodialysis patients received more transfusions ($P = .03$) than noninfected patients. There was no significant difference in liver biochemistry between GBV-C/HGV-infected and noninfected patients and between blood donors in each of the four racial groups. The high prevalence of GBV-C/HGV infection in blood donors and chronic liver disease patients, and the lack of elevated liver enzymes and clinical hepatitis in blood donors and haemodialysis patients, suggest that GBV-C/HGV may not be associated with liver disease. ***J. Med. Virol. 58:38–44, 1999.*** © 1999 Wiley-Liss, Inc.

KEY WORDS: GBV-C/HGV; blood donors; haemodialysis; chronic liver disease; racial difference; South Africa

INTRODUCTION

Liver disease is a major cause of morbidity and mortality among patients with end-stage renal disease due primarily to hepatitis B virus (HBV), hepatitis C virus (HCV), alcohol, and hemosiderosis [Rao and Anderson, 1992; Huang, 1997]. However, the etiology of liver disease remains unknown in about 4–23% of dialysis patients [Rao and Anderson, 1992; Huang, 1997]. Approximately 10–20% of non-A-E hepatitis cases remain unidentified, suggesting the existence of additional viral agents [Linnen et al., 1996]. The recent identification of GB virus C (GBV-C) [Simmons et al., 1995] followed by the identification of hepatitis G virus (HGV) [Linnen et al., 1996], are among possible transfusion transmissible viruses currently being investigated for human hepatitis of unknown etiology.

Whilst some studies did not show a direct association between GBV-C/HGV infection and liver pathology [Alter et al., 1997a, 1997b], other studies have reported a spectrum of liver pathology [Mushahwar and Zuckerman, 1998]. The site of GBV-C/HGV infection and replication in the liver, peripheral blood mononuclear cells, or other tissues have been observed by some investigators [Madejon et al., 1997; Saito et al., 1997], whilst other studies suggest that the actual site of GBV-C/HGV replication remains to be identified [Laskus et al., 1997; Mellor et al., 1998]. There is controversy about the relevance of GBV-C/HGV infection

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in fulminant hepatic failure [Heringlake et al., 1996; Toshiba et al., 1996; Kanda et al., 1997].

Only some 12–15% of chronic non-ABC hepatitis patients are GBV-C/HGV infected [Linnen et al., 1996]. High prevalence of GBV-C/HGV has been found in subjects with frequent parenteral exposure and in groups at high risk of exposure to blood and blood products [Jarvis et al., 1996; Linnen et al., 1996], including intravenous drug abusers [Aikawa et al., 1996; Linnen et al., 1996], patients on maintenance haemodialysis [Masuko et al., 1996; Tsuda et al., 1996; Lampe et al., 1997; Shrestha et al., 1997; Wang et al., 1997; Murthy et al., 1998], multitransfused individuals [Linnen et al., 1996; Neilson et al., 1996], and haemophiliacs [Jarvis et al., 1996; Linnen et al., 1996; Castelling et al., 1998]. In non-African countries, the highest prevalence of GBV-C/HGV has been documented in patients with acute and chronic HCV (18%) infection [Linnen et al., 1996]. The risk of GBV-C/HGV infection seems to be increased in those co-infected with HBV or HCV [Linnen et al., 1996; Alter et al., 1997a]. These results indicate that GBV-C/HGV is transfusion transmissible, therefore patients receiving blood transfusions, haemodialysis, and organ transplant patients are at risk of infection. Most GBV-C/HGV infections remain subclinical and resolve after the loss of serum GBV-C/HGV RNA with the concomitant detection of antibodies to the putative envelope E2 protein (anti-GBV-C/HGV E2) [Dille et al., 1997; Tacke et al., 1997]. There is evidence to suggest that anti-E2 is a neutralising and protective antibody [Hassoba et al., 1998; Tillmann et al., 1998].

Very little is known about the epidemiology and pathogenic role of GBV-C/HGV in Southern Africa [Lightfoot et al., 1997; Mphahlele et al., 1997; Tucker et al., 1997; Castelling et al., 1998]. Because GBV-C/HGV is transmitted parenterally, we screened for the presence GBV-C/HGV RNA and anti-E2 in a high-risk population of haemodialysis and chronic liver disease patients and a representative sample of blood donors (from four racial groups) in the province of KwaZulu Natal, South Africa. The aim of the study was also to determine an association, if any, between GBV-C/HGV infection and liver disease.

METHODS

Patients

The study population included 70 patients with chronic renal failure who were undergoing maintenance haemodialysis and 98 consecutive patients with chronic liver disease. As a control group, 232 adult unpaid volunteer blood donors were studied from the four racial groups (Africans, Indians, Whites, and "Coloureds" [persons of mixed origin]). Patients gave written, informed consent, and the study was approved by the Ethics Committees of the University and the Natal and Blood Transfusion Services.

Serological Assays

Sera were tested for serological markers for HBV (HBsAg, HBeAg) by commercial radioimmunoassay (RIA; Abbott Laboratories, North Chicago, IL). Antibodies to the putative envelope protein (E2) of GBV-C/HGV were determined by enzyme-linked immunosorbent assay (ELISA) "μ Plate Anti-HG_{env}" (Boehringer Mannheim, Germany) according to the manufacturer's instructions. Liver enzymes and liver function tests were carried out by routine multianalysis systems in the central laboratory.

RNA Extraction and Reverse Transcription

HCV and GBV-C/HGV viral RNA were extracted and purified from serum with a QIAmp Viral RNA Kit (Qiagen, Gmbh, Germany). Complementary DNA (cDNA) was synthesized from 10 μl of extracted RNA melted at 70°C for 10 min and added to 15 μl of a mix containing 1.5 μM $pd(N)_6$ random hexamers (Pharmacia Biotech, Uppsala, Sweden), 5× first strand buffer (50 mM Tris-HCl [pH 8.3]; 40 mM KCl; 6 mM $MgCl_2$; 1 mM DTT) (GibcoBRL, Technologies, Paisley, UK); 250 U M-MLV reverse transcriptase (GibcoBRL), 0.5 mM dNTPs (Promega, Madison, WI), and diethylpyrocarbonate treated-H_2O and incubated at 37°C for 1 hr. The reaction was terminated by incubation at 95°C for 10 min.

HCV RNA Detection

The presence of HCV RNA in serum was determined by reverse-transcription nested polymerase chain reaction (RT-PCR) using primers from the 5′ NCR region, as described previously [Brown et al., 1992] .

GBV-C/HGV RNA Detection

Highly conserved nested primers using published sequences from the 5′ NCR of GBV-C/HGV genomes were used to carry out the nested PCR [Jarvis et al., 1996]. Briefly, 5 μl of the cDNA were added to 95 μl of a first round PCR mix containing 0.2 mM dNTPs (Promega); l0× PCR buffer (1.5 mM $MgCl_2$, 20 mM $[NH_4]_2SO_4$; 75 mM Tris-HCl [pH 9.0]; 0.01% [w/v] Tween) (Advanced Biotechnologies, Surrey, UK); diethylpyrocarbonate (DEPC)-H_2O, 2.5 U Thermoprine ^{plus} DNA Polymerase (Advanced Biotechnologies) and 50 pmol of each primer located at positions 108 (5′-AGGTGGTGGATGGGTGAT-3′; sense, outer) and 531 (5′-TGCCACCCGCCCTCACCCGAA-3′; antisense, outer) (Oswald DNA Services, Southampton, UK). Two microliters of the first-round PCR product were added to 48 μl of PCR mix containing 0.2 mM dNTPs (Promega); 10× PCR buffer; DEPC-H_2O, 2.5 U Thermoprine ^{plus} DNA Polymerase, and 50 pmol/μl of each primer located at positions 134 (5′-TGGTAGGTCGTAAATCCCGGT-3′; sense, inner) and 476 (5′-TGRGCTGGGTGGCCYCATGCWT-3′; antisense, inner). Amplification was over 25 cycles for both first and second rounds for PCR, using the following temperatures: 96°C 1 min, 50°C 1 min, 72°C 2.5 min, with a final extension at 72°C 10 min followed by

TABLE I. Demographic and Biochemical Features in Blood Donors (n = 232) With and Without GBV-C/HGV Infection

Blood donors	GBV-C/HGV$^+$ Mean ± SD (Range)	GBV-C/HGV$^-$ Mean ± SD (Range)	P
African (n = 76)	(n = 29)	(n = 47)	
ALT (U/L)	17.24 ± 11.40 (9–69)	15.68 ± 6.6 (5–45)	NS
Age (years)	32.10 ± 10.06 (19–48)	28.38 ± 10.53 (18–59)	NS
Sex (M:F)	8.6:1	4.2:1	NS
Asian (n = 52)	(n = 2)	(n = 50)	
ALT (U/L)	16.5 ± 2.12 (15–18)	23.48 ± 16.82 (7–92)	NS
Age (years)	47.0 ± 7.07 (42–52)	30.68 ± 12.09 (17–67)	NS
Sex (M:F)	1:1	11.5:1	NS
White (n = 49)	(n = 11)	(n = 38)	
ALT (U/L)	24.45 ± 11.64 (11–54)	23.76 ± 13.83 (8–63)	NS
Age (years)	40.27 ± 9.71 (19–53)	39.32 ± 15.19 (17–67)	NS
Sex (M:F)	2:0	2.25:1	NS
Coloured (n = 55)	(n = 2)	(n = 53)	
ALT (U/L)	12.5 ± 2.12 (11–14)	19.06 ± 12.51 (5–73)	NS
Age (years)	19.0 ± 1.41 (18–20)	31.23 ± 12.46 (17–68)	NS
Sex (M:F)	2.67:1	1.9:1	NS

GBV-C, GB virus C; HGV, hepatitis G virus; ALT, alanine aminotransferase; NS, not significant. Coloured, persons of mixed origin.

a 4°C soak. Known positive and negative GBV-C/HGV sera together with DEPC-H_2O were included in each run. The 344-bp PCR product was detected in 2% agarose gel prepared in 1× TBE buffer stained with ethidium bromide (10 mg/ml).

Statistical Analysis

Blood donor, haemodialysis, and chronic liver disease patients' data were analysed separately. The chi-square test was used to compare groups (GBV-C/HGV positive vs. GBV-C/HGV negative) with respect to categorical data, and Student's t-test was applied to continuous data. Analysis of covariance was used to compare GBV-C/HGV-positive and GBV-C/HGV-negative patients with respect to liver function test, adjusting for age, sex, and disease group. Multiple logistic regression was used to determine any confounding influence of age, sex, and disease group on GBV-C/HGV. Blood donors were compared with patients with chronic liver disease and haemodialysis patients with regard to demographic parameters using Student's t-test or the chi-square test.

RESULTS

GBV-C/HGV RNA

Of the 232 unpaid volunteer blood donors 76 (32.8%) were African, 49 (21.1%) Whites, 52 (22.4%) Asians, and 55 (23.7%) Coloureds. All the blood donors were negative for HCV, HBV, and human immunodeficiency virus (HIV). None of the donors had a history or clinical evidence of liver disease. Overall, GBV-C/HGV RNA was detected in 44 of 232 (18.9%) blood donors (29/76 [38.2%] African, 2/52 [3.8%] Asian, 2/55 [3.6%] Coloured, and 11/49 [22.4%] White) (Table I). There was no significant difference in the prevalence of GBV-C/HGV RNA between Asian and Coloured blood donors (P = .81) and between African and White blood donors (P = .07). However, there was a significant difference in the prevalence of GBV-C/HGV RNA between the other racial groups (Asians vs. Whites [P < .005]; Asians vs. Africans [P < .00001; Coloureds vs. Whites [P < .005) and Coloureds vs. Africans [P < .00001]).

Among the 70 haemodialysis patients, there were 30 (42.9%) Africans, 29 (42.4%) Asians, 3 (4.3%) Coloureds, and 8 (11.4%) Whites. Seventeen (17) of 70 (24.3%) haemodialysis patients (5/30 [16.7%] Africans; 8/29 [27.6%] Asians; 1/3 [33%] Coloureds; and 3/8 [11.43%] Whites) were infected with GBV-C/HGV (95% confidence interval [CI] = 5.7%; 18.7%). GBV-C/HGV-positive patients tended to have a longer duration of dialysis (not significant) and have had more transfusions (P = .03) than noninfected patients (Table II). Four of 70 patients (5.7%) were infected with HCV but only 2/17 (11.76%) GBV-C/HGV-positive patients were co-infected with HCV.

Most of the patients with chronic liver disease were Black [88 Africans (90%), 7 Asians (7%), 2 Coloureds (2%), and 1 White (1%)]. The diagnosis of chronic liver disease was confirmed by histology in 81 patients and by peritoneoscopy alone in 17 patients. The etiology of chronic liver disease in most of the patients was due to alcohol abuse, viral infection, or a combination of the two. In 5 patients, chronic liver disease was due to autoimmune liver disease and in 7 it was cryptogenic. None of the patients gave a history of blood transfusions or intravenous drug abuse. The prevalence of GBV-C/HGV RNA in patients with chronic liver disease was 12.2% (12/98) (95% CI = 5.7%; 18.7%) (Table II). Only 2 of 10 patients with HBV were co-infected with GBV-C/HGV. GBV-C/HGV RNA was detected more frequently in patients with an alcoholic etiology alone (6/56 [11%]) or in combination with alcohol and HBV (1/10 [10%]) or alcohol and HCV (1/4 [25%]). Two of seven (29%) patients with cryptogenic liver disease were GBV-C/HGV RNA positive.

There was no significant difference in the prevalence

TABLE II. Demographic and Biochemical Features of Chronic Liver Disease and Haemodialysis Patients With and Without GBV-C/HGV Infection

	GBV-C/HGV$^+$ Mean ± SD (Range)	GBV-C/HGV$^-$ Mean ± SD (Range)	*P*
Chronic liver disease			
(n = 98)	(n = 12)	(n = 86)	
Age (years)	45.7 ± 15.9 (18–75)	44.8 ± 16.8 (2–81)	NS
Sex (M:F)	1.4:1	1.2:1	NS
T-Bil (umol/L)	68.6 ± 73.4 (7–235)	63.3 ± 83.3 (7–506)	NS
ALP (U/L)	197.0 ± 163.29 (18–576)	173.8 ± 143.41 (3–838)	NS
AST (U/L)	92.2 ± 48.4 (23–173)	154.6 ± 350.6 (14–2454)	NS
Haemodialysis (n = 70)	(n = 17)	(n = 53)	
Age (years)	38.17 ± 12.06 (17–65)	39.57 ± 12.76 (15–67)	NS
Sex (M:F)	1:0.8	1:0.8	NS
Dialysis (months)	76.23 ± 50.90 (12–180)	59.60 ± 54.82 (3–216)	NS
TRANSF (no.)	12.58 ± 7.79 (5–31)	8.07 ± 7.17 (2–30)	.03
AST (U/L)	14.88 ± 5.33 (6–28)	18.60 ± 13.22 (6–90)	NS
ALT (U/L)	12.58 ± 6.97 (5–32)	17.92 ± 18.24 (4–101)	NS
ALP (U/L)	132.58 ± 146.39 (36–667)	107.11 ± 131.50 (5–920)	NS
T-BIL (umol/L)	11.94 ± 2.22 (8–17)	11.32 ± 2.25 (5–17)	NS

GBV-C, GB virus C; HGV, hepatitis G virus; TRANSF, transfusion; AST, aspartate aminotransferase; ALT, alanine aminotransferase; ALP, alkaline phosphatase; T-BIL, total bilirubin; NS, not significant.

of GBV-C/HGV RNA between blood donors and patients. There was a significant difference in the prevalence of GBV-C/HGV RNA between with chronic liver disease (12/98; 12.2%) and haemodialysis patients (17/70; 24.2%) (P = .04).

Anti-E2

There was a higher prevalence of anti-E2 in African blood donors (21/76; 27.6%) than White (14/49; 8.2%), Coloured (7/55; 12.7%), or Asian (3/52; 5.7%) blood donors (Table III), and this difference was significant (P < .05). There was a significant difference (P < .05) in the prevalence of anti-E2 in chronic liver disease (32/98 [32.7%]), and haemodialysis patients (18/70 [25.7%]) compared with blood donors (35/232 [15.1%]) (Table III). There was no significant difference in anti-E2 seroprevalence between chronic liver disease and haemodialysis patients (P = .33).

African blood donors (45/76; 59.2%) had a greater exposure to GBV-C/HGV infection (RNA and/or anti-E2 positive) compared with White (15/49; 30.6%; P = .002), Coloured (9/55; 16.4%; P < .00001), and Asian (5/52; 9.6%; P < .00001) blood donors. There was a significant difference (P < .05) in the overall exposure to GBV-C/HGV (RNA and/or anti-E2 positive) between blood donors (31.9% [74/232]) and patients (43.9% [49/98] vs. 47.1% [33/70]), respectively (Table III). GBV-C/HGV RNA and anti-E2 were mutually exclusive in almost all (88–100%) patients and blood donors. GBV-C/HGV RNA and anti-E2 was positive simultaneously in only 1/98 (1.1%) patients with chronic liver disease, 2/70 (2.9%) haemodialysis patients, and in 5/232 (2.2%) blood donors.

Liver Biochemistry

Elevated alanine aminotransferase (ALT) levels were observed in only 2/70 (2.85%) dialysis patients. Mildly elevated ALT levels were observed in 6/232 (2.6%) blood donors. Only 1/44(2.3%) GBV-C/HGV-positive donors had a mild elevated ALT level (69 U/L). Using multivariate analysis and controlling for the influence of confounding factors (age, sex, and disease groups), no significant differences were observed between GBV-C/HGV infected and noninfected blood donors in each of the four racial groups (Table I), nor were such differences observed in patients with chronic liver disease and haemodialysis patients (Table II).

DISCUSSION

Although the study population may not be representative of the overall prevalence of GBV-C/HGV in KwaZulu Natal, the results indicate that there is a high prevalence of GBV-C/HGV infection in KwaZulu Natal, South Africa and that there are racial differences in the prevalence of GBV-C/HGV infection. The overall prevalence of GBV-C/HGV RNA in blood donors in the province of KwaZulu Natal (18.9%) is higher than that of blood donors in Gauteng (11.1%) [Castelling et al., 1998], with the prevalence being higher among African (38%) and White (22%) blood donors in KwaZulu Natal compared with African (29%) and White (8,5%) blood donors in Gauteng [Castelling et al., 1998]. Tucker et al. [1997] reported a GBV-C/HGV RNA prevalence of 6.3% in a predominantly Coloured rural community in the Eastern Cape that was higher than the 4% of Coloured blood donors infected with GBV-C/HGV in KwaZulu Natal. However, volunteer blood donors represent a healthier population than rural communities.

The prevalence of anti-E2 in healthy blood donors ranges from 2.7% to 20.3% in different parts of the world [Dille et al., 1997; Tacke et al., 1997; Ross et al., 1998]. The overall anti-E2 prevalence in the blood donor population (14.9%) was lower than the 20.3% reported by Ross et al. [1998] for South African blood donors. However, Ross et al. [1998] did not define the racial breakdown of the blood donor population, and our results indicate a racial difference in anti-E2 prevalence. The lower prevalence of viraemia (3.8%)

TABLE III. GBV-C/HGV RNA and Anti-E2 Seroprevalence Among Blood Donors and Patients on Maintenance Haemodialysis or With Chronic Liver Disease

Study population	No.	GBV-C/HGV RNA$^+$ (%)	GBV-C/HGV anti-E2$^+$ (%)	GBV-C/HGV RNA and anti-E2$^+$ (%)	GBV-C/HGV exposure RNA and/or anti-E2$^+$ (%)
Blood donors					
African	76	29 (38.2)	21 (27.6)	5 (6.5)	45 (59.2)
White	49	11 (22.4)	4 (8.2)	0 (0)	15 (30.6)
Asian	52	2 (3.8)	3 (5.7)	0 (0)	5 (9.6)
Coloured	55	2 (3.6)	7 (12.7)	0 (0)	9 (16.4)
Total	232	44 (18.9)	35 (15.1)	5 (2.1)	74 (31.9)
Chronic liver disease	98	12 (12.2)	32 (32.7)	1 (1.0%)	43 (43.9)
Haemodialysis	70	17 (24.3)	18 (25.7)	1 (2.9)	33 (47.1)

GBV-C, GB virus C; HGV, hepatitis G virus; Coloured, persons of mixed origin.

and anti-E2 response (5.7%) in the Asian blood donors compared with other racial groups is in agreement with the low prevalence of anti-E2 (2.7–6.3%) in Asian countries [Ross et al., 1998] and reflects the infrequent exposure to GBV-C/HGV in this community. Anti-E2 prevalence in potential risk groups was higher (25.7% vs. 32.7%) than in blood donors (15.1%) ($P < .05$). Concurrent detection of viraemia and seropositivity was seen in only 2–3% of our study population. This low rate probably indicates an overlap between E2 seroconversion and presence of viraemia [Dille et al., 1997; Tacke et al., 1997]. It may be possible that at the time of testing these patients were still viraemic and in the process of seroconversion. Viraemia and seroreactivity was almost mutually exclusive. The combined overall exposure of GBV-C/HGV infection in African blood donors (59.2%) was higher compared with 46.7% (PCR + anti-E2) reported in a West African population [Dille et al., 1997]. GBV-C/HGV infection appears to be a common infection in our community.

There is a higher prevalence of GBV-C/HGV infection in apparently healthy individuals in African countries (10–40%) [Dawson et al., 1996; Dille et al., 1997; Mphahlele et al., 1997; Tucker et al., 1997; Castelling et al., 1998], with infection occurring more commonly during childhood [Mphahlele et al., 1997], compared with blood donor groups in non-African countries (1–4%) [Linnen et al., 1996; Masuko et al., 1996; Moaven et al., 1996; Alter et al., 1997b; Roth et al., 1997; Wang et al., 1997]. The reason for the high prevalence of GBV-C/HGV in blood donors worldwide and basis for the racial differences in GBV-C/HGV infection in the South African blood donor populations is not known. Racial differences in the prevalence of HAV [Sathar et al., 1994], HBV [Dusheiko et al., 1989], and HCV [Soni et al., 1993] infections due to differences in socioeconomic factors in South Africa is well documented. Whether this difference holds true for GBV-C/HGV is not known, although a relationship was noted between GBV-C/HGV infection and the lack of waterborne sewage [Tucker et al., 1997]. The differences in the prevalence of detecting GBV-C/HGV infection may be due to the differences in the sensitivity of the various PCR protocols and primers (derived from various regions of the genome) used by various investigators. The nested PCR used in this study is likely to contribute to an increase in the sensitivity of the assay as compared with the one step PCR procedures. In addition the testing of anti-E2 greatly extends the ability of RT-PCR to define the epidemiology of GBV-C/HGV.

In this study, most haemodialysis patients and blood donors, including those with GBV-C/HGV infection, had normal liver enzymes. The high prevalence of GBV-C/HGV in blood donors and haemodialysis patients without biochemical evidence of liver damage suggests that many patients are viraemic in the absence of liver disease. However, normal ALT levels do not prove definitely normal histology, as shown for HCV infection [Alberti et al., 1991]. No significant difference was observed between GBV-C/HGV RNA-positive and -negative patients. More recently, no association was reported between GBV-C/HGV and hepatocellular carcinoma in Black South Africans [Lightfoot et al., 1997]. The prevalence of HCV in haemodialysis patients (4/70; 5.7%) and in patients with chronic liver disease (6/98; 6.1%) in KwaZulu Natal is similar to that reported previously (4.8%) and confirms the low prevalence of HCV [Soni et al., 1993].

In non-African countries, the mode of transmission of GBV-C/HGV occurs parenterally and is considered to be similar to HCV [Linnen et al., 1996]. In this study, GBV-C/HGV-infected dialysis patients tended to have had more transfusions and a longer duration of dialysis than noninfected patients. Therefore, most patients on maintenance haemodialysis acquired their GBV-C/HGV infection through the transfusions they received [De Lamballeric et al., 1996; Masuko et al., 1996; Tsuda et al., 1996; Lampe et al., 1997; Shrestha et al., 1997; Wang et al., 1997; Castelling et al., 1998; Murthy et al., 1998]. The absence of a history of blood transfusions and intravenous drug abuse in patients with chronic liver disease, in addition to the high prevalence of GBV-C/HGV in blood donors and in rural communities, especially in African children, tends to suggest that GBV-C/HGV is being transmitted by as yet undefined nonparenteral routes in South Africa. Both vertical [Feucht et al., 1996] and sexual [Ibanez et al., 1998; Scallan et al., 1998] transmission of GBV-C/HGV has been suggested. HBV infection is endemic amongst the African population of South Africa and the risk of horizontal transmission of HBV is well recognised [Dusheiko et al., 1989]. Whether the association of

GBV-C/HGV with HBV implies that GBV-C/HGV might spread by sexual or prenatal routes cannot be determined by this study and would require a more comprehensive epidemiological study to elucidate the high prevalence of GBV-C/HGV in our population.

Although histochemical and hybridisation studies of liver biopsies to detect GBV-C/HGV were not undertaken in this study, the higher prevalence of GBV-C/HGV in blood donors and patients with chronic liver disease and the lack of elevation in liver enzymes as well as clinical hepatitis in blood donors and haemodialysis patients would suggest that GBV-C/HGV may not be associated with liver disease.

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